Unraveling the Mystery of Goldblatt Hypertension

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In hypertension caused by unilateral renal artery stenosis, the nonstenotic kidney becomes renin depleted but fails to prevent hypertension. The nonstenotic kidney mysteriously develops elevated intrarenal angiotensin II (ANG II) content. Rats chronically infused with ANG II exhibit a similar hypertensive process. The augmentation of intrarenal ANG II is due to receptor-mediated internalization and continued ANG II formation, which provide a hypertensinogenic stimulus.

Although there have been great strides in the development of antihypertensive drugs, hypertension continues to be a major health problem affecting over 20% of the adult population. If left untreated, hypertension causes damage to the vascular endothelium, resulting in a proliferative response, arteriosclerosis, and consequent end-organ damage leading to increased risks of stroke, coronary arterial disease, myocardial infarction, congestive heart failure, and chronic renal failure (10). Although hypertension is often regarded as the cause of renal disease and chronic renal failure, it is also recognized that hypertension may be the consequence of defects in renal microcircularity and/or tubular transport function that compromise the normal capability of the kidney to maintain sodium balance at normal arterial pressures. Even when there is no primary intrarenal derangement, an impairment in renal function secondary to inappropriate humoral or neural stimulation to the kidney may exist. In essence, a widely held premise is that hypertension cannot coexist in the presence of normal kidney function (3, 6, 9).

Responses to unilateral renal arterial stenosis

One very intriguing experimental model of hypertension is the two-kidney, one-clip (2K1C) Goldblatt model (4) in which hypertension is induced by unilateral stenosis of the renal artery (2, 7, 8, 11). The clip is not severe enough to cause ischemia; however, the reduced renal perfusion pressure stimulates increased renin synthesis and release from the clipped kidney. As shown in Fig. 1, renin enzymatically cleaves angiotensin I (ANG I) from angiotensinogen, and angiotensin-converting enzyme (ACE) acts on ANG I to produce angiotensin II (ANG II). Circulating ANG II, via its direct vascular effects, acutely increases total peripheral resistance and...
raises blood pressure but also has actions on almost every organ system (9). Many of these actions contribute to increases in peripheral vascular resistance and arterial pressure and also to long-term proliferative responses (Fig. 1B). With the progressive increases in systemic arterial pressure, perfusion pressure and flow to the clipped kidney are restored. The nonclipped contralateral kidney, subjected to progressive elevations in arterial pressure, becomes renin depleted and would be expected to increase sodium excretion due to the phenomenon of pressure natriuresis (3,6). Because one normal kidney is sufficient to maintain fluid and sodium balance.
at normal arterial pressures, the mystery of Goldblatt hypertension resides in the inability of the presumably normal kidney to prevent the development of hypertension. In this setting, it is apparent that the normal nonclipped kidney is clearly not the causative factor, yet it fails to elicit the appropriate natriuretic response to the developing hypertension (2, 3, 6, 7, 11).

As depicted in Fig. 2, the cascade of events initiated by the increased renin secretion from the clipped kidney leads to increases in circulating angiotensin II (ANG II), thus leading to various ANG II-mediated changes as shown in Fig. 1. As explained in the text, ANG II content in the renin-depleted nonstenotic kidney also increases through mechanisms not dependent on renin. ACE, angiotensin-converting enzyme; A, angiotensinogen; N/C, no change.

Activation of the renin-angiotensin system

The direct and indirect effects of the increased circulating ANG II concentrations along with the resultant increases in aldosterone production and the ANG II-dependent increases in the activity of the sympathetic nervous system contribute to the impaired excretory capability of the nonclipped kidney (9, 11). These interacting effects contribute greatly to the early developmental stages of 2K1C Goldblatt hypertension, when plasma renin activity and circulating ANG II concentrations are elevated. As renal perfusion pressure to the clipped kidney is reestablished, however, the plasma renin activity and circulating ANG II concentrations return toward the normal range, whereas the arterial pressure remains elevated (5). Even during this maintenance stage, ANG II continues to exert powerful effects on function of the nonstenotic kidney (2, 7, 11, 13). Many studies have shown that the nonclipped kidney is highly responsive to pharmacological blockade of the renin-angiotensin system (7–9, 11). In the example shown in Fig. 3, ACE inhibition increased renal blood flow, glomerular filtration rate (GFR), and sodium excretion even as arterial pressure was reduced (7). The effects of the small increase in GFR are compounded by the associated decreases in proximal tubular reabsorption rate that cascade further into proportionally greater increases in distal volume delivery and eventually to much greater relative increases in sodium excretion. When ACE inhibitors or ANG II receptor antagonists were given chronically to 2K1C Goldblatt rats, the hypertension and the ANG II-induced decreases in renal function of the nonclipped kidneys were prevented. Renal blood flow and GFR were increased in the nonclipped kidneys of the clipped rats treated chronically with these blockers (8).

Intrarenal ANG II levels

The finding that the renin-angiotensin system continues to exert a powerful influence on the nonclipped kidney even after the circulating ANG II concentrations return to normal suggested that intrarenal ANG II levels remain elevated even in the face of suppression of renin formation. To determine if the intrarenal ANG II contents of the nonclipped kidneys were dissociated from the circulating ANG II levels, we performed a detailed evaluation of the intrarenal ANG I and ANG II contents (5). As shown in Fig. 4, the plasma ANG II levels were elevated at 7 days but were close to control values 3 wk after clipping. However, the nonclipped kidneys did not have diminished intrarenal levels of ANG II; rather, they had elevated levels of ANG II during both the early developmental stages of hypertension and even after the hypertension was well established and the plasma ANG II concentra-
tions had returned toward normal (5). Furthermore, the ANG II levels in the nonclipped kidneys were substantially greater than could be explained from the circulating plasma ANG II concentrations, making it unlikely that the renal ANG II levels were due simply to ANG II present in the trapped plasma and interstitial or tubular fluid within the kidney. The elevated intrarenal ANG II contents were also associated with increased ACE activity, indicating more efficient conversion of ANG II from ANG I (5). These data led to two important conclusions that provided clues for solving the mystery of why the nonclipped kidney fails to protect against hypertension. First, under these conditions of unilateral renal arterial stenosis, there was a clear dissociation between the renal renin content and the renal ANG II content in the nonclipped kidney. This finding indicates that renal renin depletion does not necessarily reflect ANG II depletion and raises the possibility that other “low renin” models of hypertension may also have high intrarenal ANG II levels that are not predictable from renin measurements. Second, the elevated intrarenal ANG II levels in the nonclipped kidney indicated that there was accumulation of circulating ANG II in the kidney or sustained intrarenal ANG II production through a mechanism not dependent

FIGURE 3. Segmental volume flow and tubular reabsorptive responses to angiotensin-converting enzyme (ACE) inhibition in nonclipped kidney of Goldblatt hypertensive rats. C, values obtained during control conditions in hypertensive rats; I, values observed after administration of angiotensin-converting enzyme (ACE) inhibitor. BP, blood pressure. Cascading effects on tubular volume reabsorption are reflected in the percentile changes indicated. [Data are from Huang et al. (7).]

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FIGURE 4. Plasma and kidney angiotensin II (ANG II) levels in 2-kidney, 1-clip (2K1C) Goldblatt hypertensive rats and ANG II-infused rats. See text for further details. [Data are from Guan et al. (5) and Zou et al. (15).]
on renin (5).

Functional consequences of elevated intrarenal ANG II levels

These results indicated that elevated intrarenal ANG II levels in the renin-depleted, nonclipped kidney are responsible for the altered tubular reabsorptive and hemodynamic function. Increases in intrarenal ANG II, effected either through increased delivery from the circulation or as a consequence of conversion of ANG I generated locally, directly stimulate proximal tubular reabsorptive function by enhancing the activity of the luminal membrane Na+/H+ exchanger and the basolateral membrane Na+/HCO₃⁻ cotransporter (9). These findings, together with the observation that ACE inhibition decreases proximal reabsorption rate in the nonclipped kidney (7), indicate that the elevated ANG II levels in the nonclipped kidney augment proximal reabsorption rate. More recent studies also indicate that ANG II influences distal tubule reabsorption rate as well.

One consequence of enhanced proximal reabsorption rate is a reduction in end proximal fluid flow, which causes a decrease in fluid delivery to the macula densa segment. A decrease in fluid delivery to the macula densa segment would elicit a tubuloglomerular feedback (TGF)-mediated decrease in pregglomerular vascular resistance and an increase in single nephron glomerular filtration rate of sufficient magnitude to offset or counteract the ANG II-mediated increase in proximal tubular reabsorption rate. Consequently, an ANG II-mediated increase in proximal tubular reabsorption rate, by itself, would not elicit sustained decreases in distal nephron fluid delivery and sodium excretion because of the compensatory action of the TGF mechanism to restore distal nephron volume delivery back toward control levels.

As shown in Fig. 5, ANG II also exerts a powerful modulatory influence on the overall sensitivity of the TGF mechanism (9). Administration of AT₁ ANG II receptor antagonists, such as losartan, or ACE inhibitors markedly attenuates TGF responses to increases in distal nephron perfusion rate (2, 9). Infusion of exogenous ANG II during conditions of converting enzyme blockade results in partial restoration of feedback responsiveness. Furthermore, TGF responsiveness in normal rats is enhanced by ANG II. Peritubular capillary infusions of either ANG I or ANG II, at doses that do not directly alter glomerular dynamics or elicit systemic effects, enhance the sensitivity of the TGF mechanism (9). Such an effect of ANG II to enhance TGF responsiveness shifts the operating point of the system and thereby allows GFR to be maintained at the lower distal nephron volume delivery. Thus, during conditions of elevated intrarenal ANG II levels, the interactive effects of ANG II to enhance both proximal tubular reabsorption rate and the sensitivity of the TGF mechanism are responsible for the sustained decreases in distal nephron volume delivery. This effect of reduced distal delivery is amplified further by the increased distal tubular sodium reabsorption mediated by aldosterone and by ANG II. When these intrarenal effects of ANG II are blocked, as shown in Fig. 3, there is a marked natriuretic response as long as arterial pressure does not fall excessively (7, 9, 11).
The TGF responsiveness in the nonclipped kidney has been found to be normal or slightly enhanced compared with normotensive animals (2, 11). Administration of the nonpeptide AT₁ receptor antagonist, losartan, markedly attenuated the magnitude of the TGF responses in the nonclipped kidney, indicating that ANG II, acting via AT₁ receptors, enhances the sensitivity of the TGF mechanism in the nonclipped kidney of Goldblatt hypertensive rats (2). This augmented TGF responsiveness, combined with the direct stimulatory action of ANG II on proximal tubular reabsorptive rate, provides a powerful synergistic mechanism whereby the elevated ANG II levels impair the ability of the nonclipped kidney to maintain normal rates of sodium excretion at normotensive pressures. The elevated ANG II levels would also attenuate the natriuretic action of the nonclipped kidney to elevations in arterial pressure. In this manner, the direct renal vascular and tubular effects of ANG II play an important role in the development and maintenance of hypertension that occur as a consequence of unilateral renal arterial stenosis. When combined with the ANG II-dependent increases in aldosterone levels along with direct effects of ANG II on distal nephron transport mechanisms, the actions of ANG II cascade into an extremely powerful sodium-retaining stimulus (9).

**ANG II-infused hypertension**

The dissociation between intrarenal renin levels and ANG II content in the nonclipped kidney suggests that, under these conditions, intrarenal ANG II levels are not being regulated by renin. To evaluate mechanisms by which increases in circulating ANG II augment the intrarenal ANG II levels, we used the ANG II infusion model of hypertension, which does not have elevated circulating or tissue renin activity (12–15). Instead of clipping one kidney, we removed the kidney and substituted an osmotic minipump containing ANG II. As shown in Fig. 4, the ANG II infusion raises the plasma ANG II levels to those observed after unilateral arterial stenosis. This ANG II infusion rate does not cause immediate increases in systemic arterial pressure but rather leads to a slowly developing hypertension over a period of 6–10 days. The renal renin content and renin mRNA and the plasma renin activity are all markedly suppressed in this model (12). The critical new observation from these experiments that helped to solve the mystery was that intrarenal ANG II content also increased significantly after ~8–10 days of ANG II infusion to levels substantially greater than could be explained by the circulating ANG II. These data support the concept that modest increases in circulating ANG II elicit more substantive increases in renal ANG II content that are not dependent on renin activity. Renal function in the ANG II-infused rats is clearly influenced by the elevated ANG II levels. Blockade of AT₁ receptors with losartan led to increases in renal blood flow, GFR, and sodium excretion as well as suppression of the fractional sodium reabsorption rate (13). Furthermore, chronic administration of losartan in the drinking water during the 14 days of ANG II infusion prevented the development of hypertension (13, 15).

These results provided further clues that helped to solve the mystery. Measurements in other tissues such as the heart, aorta, and adrenal gland did not show a disproportionate increase in ANG II levels, so the changes observed seemed to be renal specific (15). It also seemed unlikely that the increases in renal ANG II were due to nonspecific accumulation because the levels expressed per gram of total kidney weight were much greater than the plasma concentrations (14, 15). However, gene regulation studies did not show an upregulation of the angiotensinogen mRNA levels and the renin mRNA levels were markedly decreased (12). The likelihood of an ANG II accumulation mechanism raised the question of whether or not the accumulation process was passive or required the mediation of ANG II receptors. In vitro studies have already shown that the ANG II receptor complex can be internalized in vascular smooth muscle cells (1). Thus further experiments were performed to determine if the intrarenal ANG II augmentation process requires AT₁ receptor activation as shown for vascular smooth muscle cells in vitro. The AT₁ receptor antagonist, losartan, was added to the drinking water of rats and, as already mentioned, prevented the development of ANG II-induced hypertension. The novel finding that helped solve the mystery was that losartan markedly reduced the intrarenal ANG II content of kidneys harvested after 13 days of ANG II infusion (15). As shown in Fig. 4, the decrease in intrarenal ANG II content occurred even though plasma ANG II concentrations increased further because of the losartan effects to stimulate renin production. This finding provided further evidence for the dissociation between renal ANG II content and circulating ANG II concentrations and, furthermore, indicated that ANG II binding to its AT₁ receptor leads to an activating mechanism responsible for increasing intrarenal ANG II levels. This AT₁-dependent mechanism increases intrarenal ANG II levels by either stimulating further intrarenal ANG II production or accumulating the circulating ANG II. Which method is correct was answered by studies infusing Val⁵-ANG II, a form...
of ANG II that can be distinguished from endogenous ANG II. Zou et al. (14) found that much of the infused Val3-ANG II was internalized by a receptor-mediated process into an intracellular compartment that protected the ANG II from degradation. Furthermore, there was a sustained production of endogenous ANG II that also contributed to the increased intrarenal ANG II contents (14).

These results have helped us to understand how a renin-depleted kidney can still have elevated ANG II levels. Although much of the circulating ANG II that binds to its receptor is internalized and degraded, some of the peptide is protected from degradation and presumably can have functional effects. The nonclipped renin-depleted kidney is clearly not depleted of ANG II. The intrarenal ANG II contents in nonclipped kidneys are almost as high as they are in the renin-rich kidney subjected to renal arterial stenosis. The functional studies indicate that the elevated ANG II levels exert significant actions to vasoconstrict the renal microvasculature, to enhance the responsiveness of the TGF mechanism, and to augment proximal and distal tubule sodium reabsorption rate through both direct and indirect mechanisms (9, 13). Activation of these synergistic actions allows the nonclipped kidney to exert a powerful hypertensinogenic influence that leads to the development and maintenance of Goldblatt hypertension. As long as the renal actions are sustained, the hypertension can be maintained even after the circulating ANG II concentrations return to the normal range.

In Goldblatt’s (4) 1934 landmark paper describing the production of hypertension by unilateral renal arterial constriction, he outlined possible mechanisms by which blood pressure could be elevated. One possibility was the accumulation or formation of a substance that effects a pressor action on the vasculature. We now know that ANG II is the hormone responsible for the elevation in blood pressure and that augmentation of intrarenal ANG II levels plays the crucial role in sustaining the hypertensinogenic process. Because both renin and renin mRNA levels in the nonclipped kidney are markedly suppressed, the elevated intrarenal ANG II levels are mediated by a mechanism not dependent on renin. On the basis of studies on ANG II-infused rats, the augmentation of intrarenal ANG II is dependent on activation of AT1 receptors and subsequent receptor-mediated internalization of ANG II and further enhancement of intrarenal ANG II formation.

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